Supplementary Information

Title: Binding of SARS-CoV-2 spike protein to ACE2 is disabled by thiol-based drugs; evidence from *in vitro* SARS-CoV-2 infection studies.

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Supplementary Figure S1

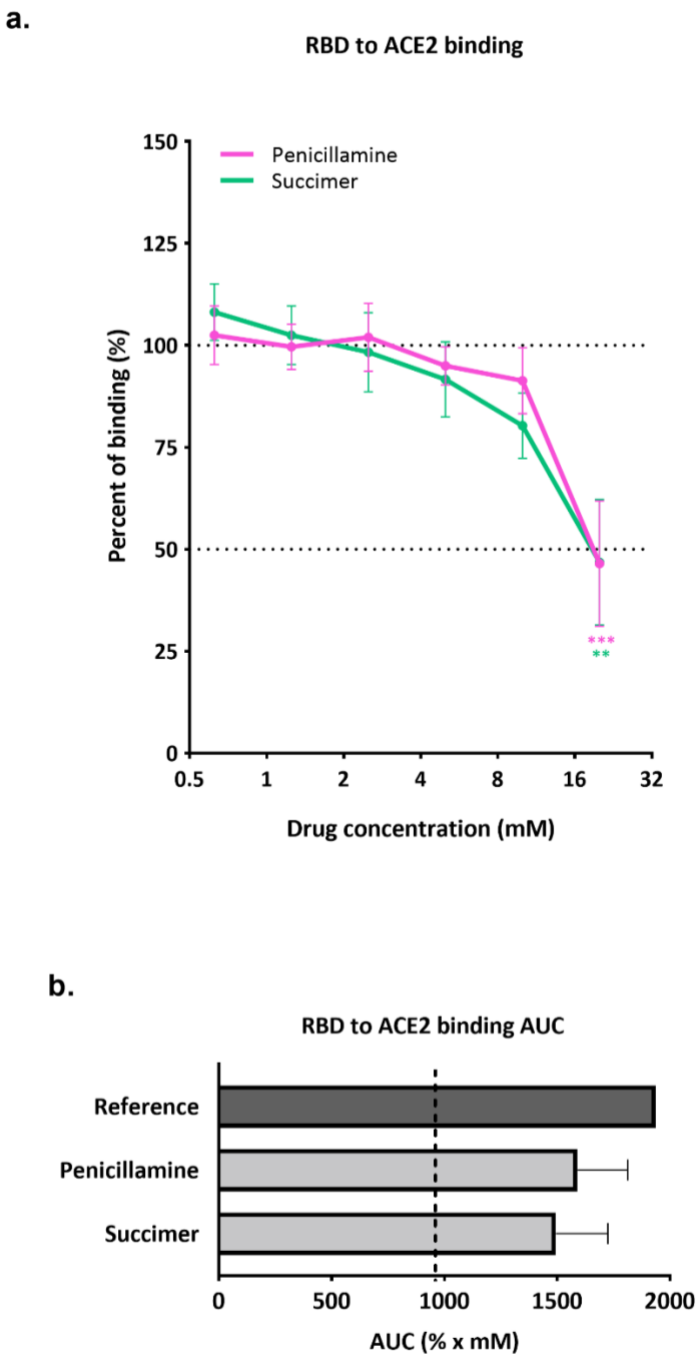

Figure S1. Effect of penicillamine and succimer on binding of SARS-CoV-2 RBD to ACE2. Panel (a) shows percent of binding in the presence of the penicillamine and succimer (n = 4 - 6). Without drug treatment, the binding was 100%, whereas treatment with the thiol-based drugs showed a decrease in the binding % relative to no drug control. The X axis is scaled to log2. Panel (b) shows area under the curve (AUC) analysis for effects of the thiol-based drugs on RBD to ACE2 binding. Reference AUC was calculated from RBD to ACE2 binding with no drug control; dashed line represents 50% of reference AUC. Data are mean ± SEM. Statistical significance was analyzed by one-way ANOVA followed by Dunnett's post-hoc analysis. Significance indicates differences from no drug control (a) or reference AUC (b). **$p \leq 0.01$, ***, $p \leq 0.001$.

Supplementary Figure S2

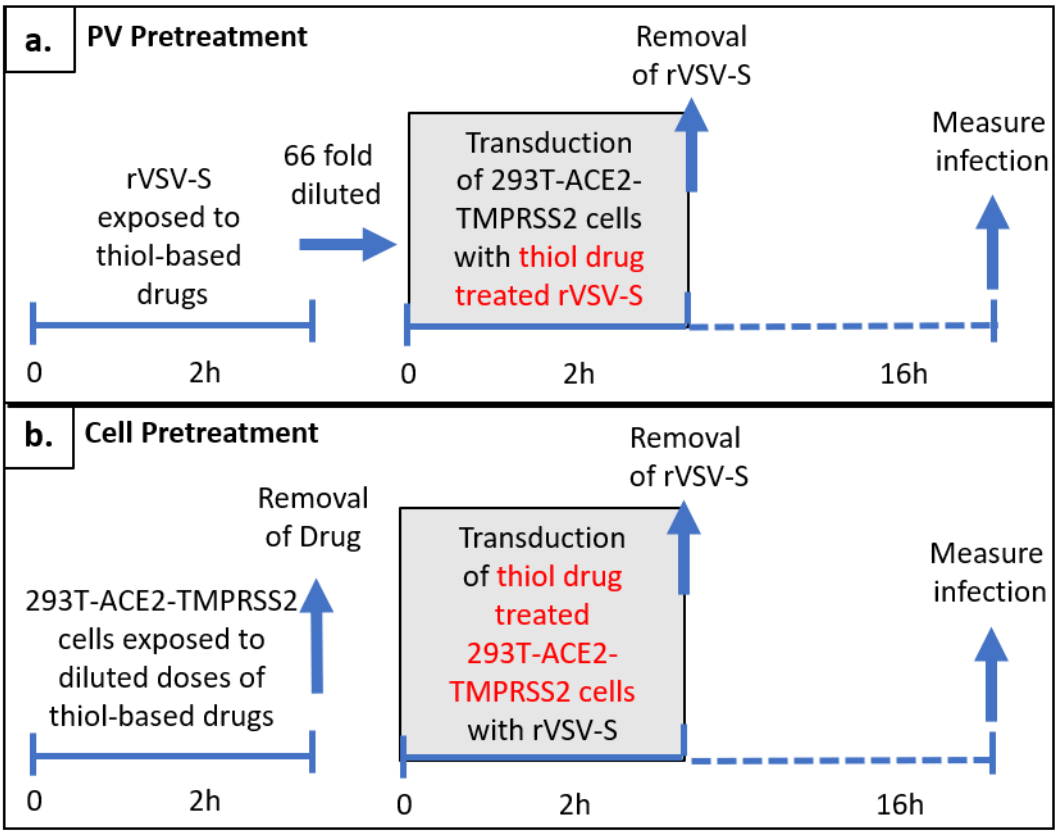

Figure S2. Schematic illustration of the strategies employed to assess thiol-based drugs as pseudovirus entry inhibitors. a. Pseudovirus (PV) pretreatment: The rVSV-S was pre-incubated with thiol-based drugs prior to transduction of the 293T-ACE2-TMPRSS2 cells. **b.** Cell pretreatment: The 293T-ACE2-TMPRSS2 cells were exposed to thiol-based drugs before the cells were transduced with rVSV-S.

Supplementary Figure S3

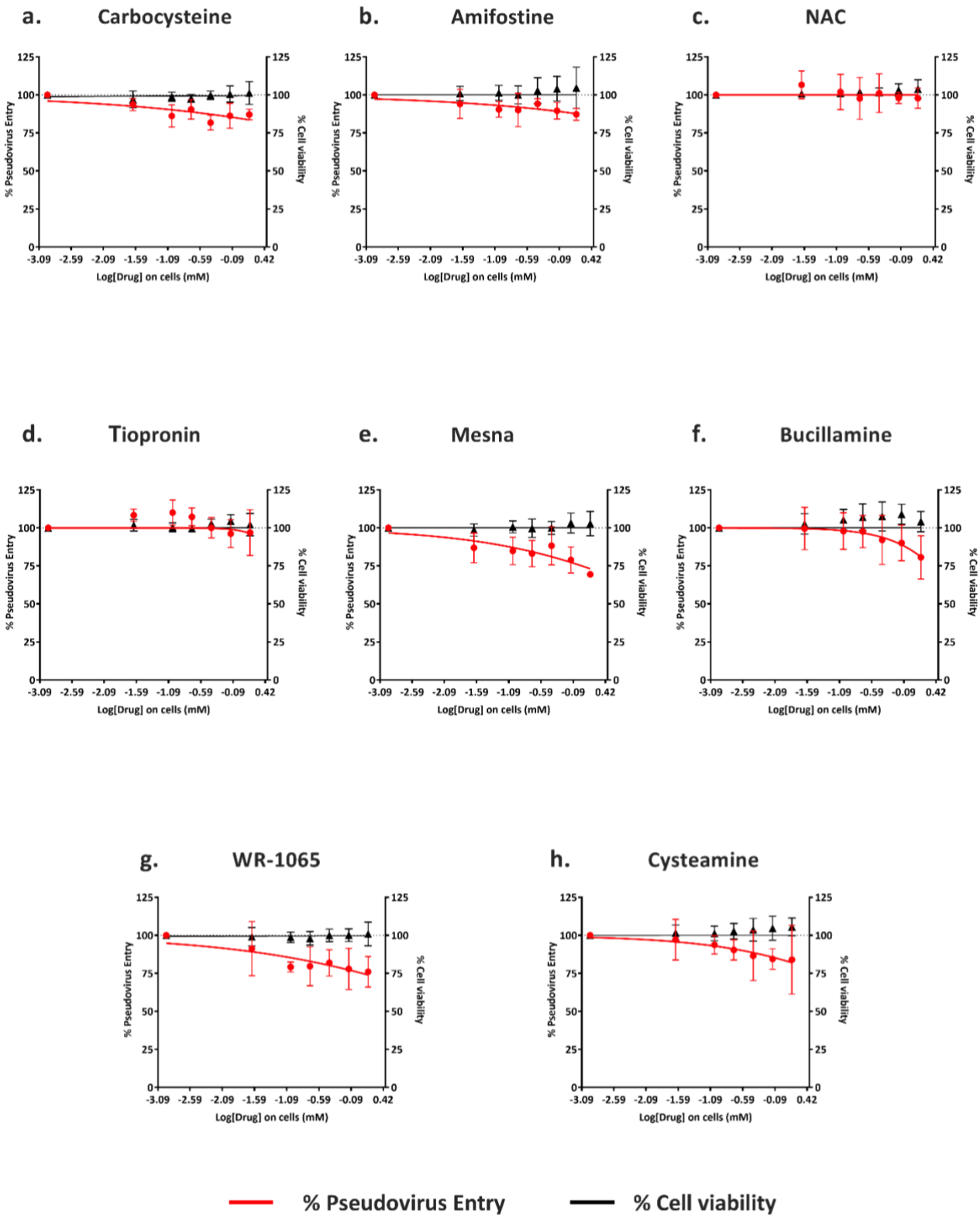

Figure S3. Effect of cell pretreatment with thiol-based drugs on the entry of pseudoviruses in the cells. Pseudovirus entry efficiency, quantified by luciferase activity, when the cells were exposed to drugs prior to transduction with untreated virus (as illustrated in Panel b of Figure S2), (n = 3). The effects of drugs on viability of 293T-ACE2-TMPRSS2 cells was quantified using Cell Titer Glo 2.0. The drug doses reflect the 66-fold dilution of drugs when pseudovirus/drug mixture was incubated with cells in the pseudovirus pretreatment strategy. The X-axis are scaled to log10. Percentage changes are with respect to no drug control which is set as 100%. Data are mean ± SD.

Supplementary Figure S4

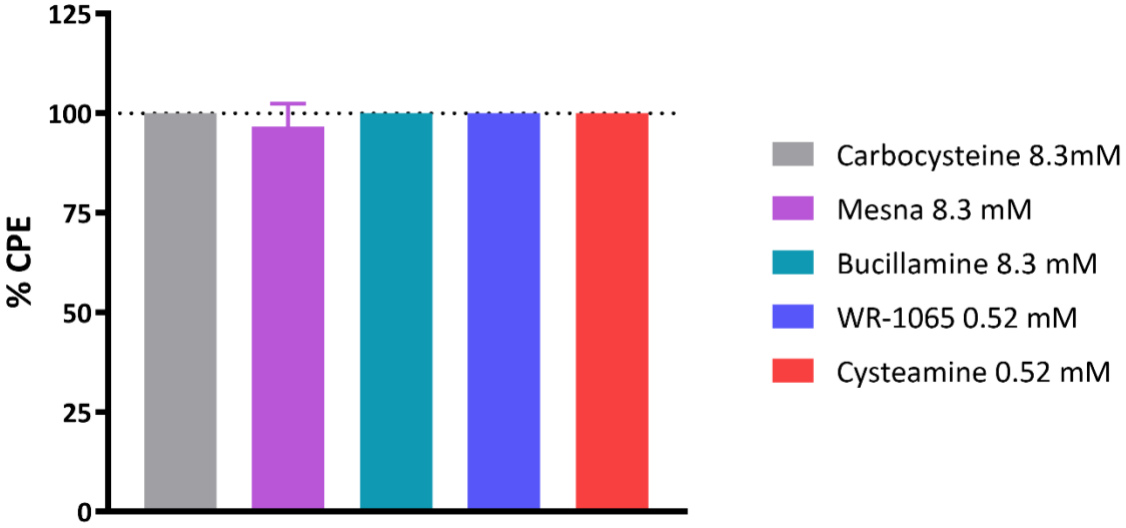

Figure S4: Effect of pretreatment of the cells with thiol-based drugs on cytopathic effects in SARS CoV2 infection. Cytopathic effects (CPE) in Vero E6 cells infected with SARS CoV-2 virus when the cells were pre-exposed to drugs prior to infection with untreated virus. A single dose of each drug was used to reflect the highest dose that the cells were exposed to in the virus pretreatment experiments. Data are mean ± SD.

Supplementary Table S1

Table S1: Comprehensive list of thiol-based drugs or drugs that generate a thiol-containing metabolite*				
	Compound	**Formulations**	**Clinical indications**	**Doses**
Monothiol drugs				
1	N-acetylcysteine	Oral, Intravenous (IV), inhaled	Mucus pathology; acetaminophen toxicity	Oral: 0.5 - 2.0 g daily IV: 300 mg/kg Inhaled: 3-5 mL of 20% solution.
2	2-mercaptoethane sulfonate, sodium salt (MESNA)	Oral, IV, inhaled (India)	Chemotherapy-related hemorrhagic cystitis	Oral: 400 mg IV: 240 mg/m^2 Inhaled: 3-6 mL of 20% solution
3	Tiopronin	Oral	Cystinuria	Oral: 1.0 g daily
4	Cysteamine	Oral, Ocular	Cystinosis	Oral: 2.0g daily
5	Amifostine	IV	Chemotherapy and radiation protective agent	IV: 200-910 mg/m^2
6	Penicillamine	Oral	Wilson's disease; Rheumatoid arthritis; heavy metal poisoning; cystinuria	Wilson's disease: 0.75- 1.5 g daily Cystinuria: 1-4 g daily Rheumatoid Arthritis: 125-250 mg daily
7	Erdosteine	Oral	Mucus pathology	600 mg daily
8	Glutathione	Oral	Supplement	50-600 mg daily
Dithiol drugs				
10	Bucillamine	Oral (in Japan and South Korea)	Rheumatoid arthritis	Oral: 300 - 600 mg/day
11	Dimercaptosuccinic acid (DMSA) (Succimer)	Oral	Heavy metal poisoning; cystinuria	10mg/kg every 8 hours for 5 days
12	2,3-Dimercaprol	Intramuscular	Heavy metal poisoning	2.5 – 12 mg/kg/day
* Not shown are three thiol containing drugs (Captopril, Zofenopril and Racecadotril) in which primary mechanism of action is not through reactions or interactions of the thiol group				